Pharmacokinetic properties of recombinant FVIIa in inherited FVII deficiency account for a large volume of distribution at steady state and a prolonged pharmacodynamic effect

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Running Title: rFVIIa PK in FVII deficiency

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Summary (241 words)

Determining the optimal frequency of replacement therapy is a clinically relevant issue in factor VII (FVII) deficiency, a bleeding disorder characterised by trace levels of a protein with a very short half-life. The main objective of this study was to clarify the discrepancy between the unfavourable PK data and positive clinical results obtained with rFVIIa. *In Vivo* Recovery (IVR) was determined in 56 patients using data from the Seven Treatment Evaluation Registry (STER) and a full PK analysis was performed in 10 severe FVII-deficient patients (FVII coagulant activity [FVII:C] <2%) in a non-bleeding state. When evaluated using FVII:C, the main study results were: i. low IVR (0.59 U dl^{-1}/U kg^{-1}), ii. a very large volume of distribution at steady state (14.72 dl kg^{-1}), iii. fast clearance (4.41 dl h kg^{-1}), iv. very short half-life (2.12 h) and mean residence time (MRT) (3.05 h). Significantly ($p < 0.001$) longer half-life (5.80 h) and MRT (8.38 hours) were obtained using a global assay, the Prothrombin Time ratio (PTr). The large volume of distribution of rFVIIa, approximately 40 times the plasma volume, the low IVR, the very short half-life and the fast clearance, indicate that a rapid and substantial diffusion towards the extravascular spaces occurs. The discrepancy between the FVII:C- and the PTr-related data indicate the presence of noticeable plasma clotting activity after rFVIIa infusion, when FVII:C is not detectable, consistent with the observation that prophylaxis is effective with less frequent administrations and low doses.

Keywords

Congenital bleeding disorder, pharmacokinetics, recombinant factor VIIa

Introduction

In congenital bleeding disorders (CBD), the mainstay for the prevention and treatment of bleeding episodes is replacement therapy (RT), the efficacy of which can be indirectly measured through pharmacokinetic (PK) methods based on the assessment of post-infusion factor levels over time (1). Specific clotting factor levels are currently considered the best determinants of haemostatic correction (1-3), and the key element for efficacy evaluation. Post-infusion clotting factor levels depend on RT dose and frequency as well as the PK profiles, which are factor specific.

Currently, little information is available on the PK of activated factor VII (FVIIa) in FVII deficiency, a CBD with an estimated prevalence of one in 500,000 individuals (4-6) and characterised by large variation in residual FVII levels and clinical phenotypes. Treatment demands for FVII deficiency may vary considerably, ranging from episodic, infrequent factor administrations to high-dose substitutions for prophylaxis or major surgical interventions. These differences in treatment demands mostly depend on the clinical severity of the defect, which may vary from a mild, impaired primary haemostasis-like clinical picture to very severe, life-threatening and invalidating forms (7, 8).

Preliminary PK data on recombinant FVIIa (rFVIIa) in patients with FVII deficiency showed a fast clearance and very short half-life (9); more recent data demonstrated an increased volume of distribution (10). Notwithstanding the unfavourable PK data reported (9, 10), rFVIIa has been successfully used for prophylaxis in patients with FVII deficiency (11-14), both in young children and adults (14). Considering this apparent discrepancy, understanding the PK profile of rFVIIa in patients with FVII deficiency is of paramount importance.

To evaluate the decline of rFVIIa in the bloodstream, we studied its PK in patients with severe FVII deficiency and compared the 'specific' FVII coagulant activity (FVII:C) assay with a 'global' test, the Prothrombin Time ratio (PTr). Further, extensive *in vivo* recovery (IVR) evaluations were performed using data collected from a large number of patients treated with rFVIIa, obtained from the Seven Treatment Evaluation Registry (STER).

Materials and methods

Patients

Two cohorts of patients received rFVIIa (NovoSeven®, Novo Nordisk A/S, Bagsværd, Denmark). The first consisted of 10 non-bleeding patients (6 females, 4 males; median age 43 years, range 9–57 years) with severe FVII deficiency (FVII:C <2%). A complete PK evaluation was performed in these patients after an rFVIIa infusion (16–24 μg kg^{-1}; mean 19.7, median 20 μg kg^{-1}). The effect of rFVIIa on FVII:C and PTr assays was evaluated pre-dose and 0.17–24 hours (0.17, 0.5, 1, 2, 4, 6, 8, 10 and 24 hours) post-dose (15) using a Non-Compartmental Analysis (NCA) (16-18). Doses of rFVIIa have been converted to IU according to the conversion factor 1 μg = 50 IU, because the FVII:C concentration was expressed as IU/dl. To analyse the decay of the PTr data, inverse values (1/PTr) were used to graphically compare the data obtained using FVII:C and PTr; values were expressed as a percentage of the highest post-infusion value (Fig. 1).

The second cohort comprised 56 patients (32 females, 24 males; median 23 years, range 2–81) from the multi-centre, prospective, observational, web-based STER (Clinicaltrials.gov identifier: NCT01269138), who required RT with rFVIIa (Table 1). Patients were selected on the basis of a cut-off of FVII:C levels of 26%, as levels higher than this threshold have an uncertain clinical relevance (19, 20). IVR was evaluated from plasma samples collected prior to and 15 minutes after RT with rFVIIa for prophylaxis, treatment of spontaneous bleeding episodes or surgical interventions. Patients with a FVII inhibitor were excluded from analyses. Only data obtained

using high-sensitivity thromboplastins (international sensitivity index [ISI] ≅1) were included in the IVR analyses to eliminate the influence of the thromboplastin ISI on the clotting assays.

In vivo recovery

Incremental IVR was calculated as the ratio between the post-infusion gain (15 minutes) of FVII and the amount of factor administered according to the following formula:

$$IVR = \frac{MPIL - BL}{TDA}$$

*w*here:

IVR is expressed as U dl^{-1}/U kg^{-1}, MPIL is maximum post-infusion level (U dl^{-1}), BL is baseline level (U dl^{-1}) and TDA is total dose administered (U kg^{-1}).

Statistical analysis

All NCA parameters were calculated using WinNonlin (Pharsight, Cary, NC, USA). Mean and standard deviation were used as position parameters to describe the variable distribution. Multivariate analysis was performed by running a generalised linear model with conditional gamma distribution and canonical link (21). This model was chosen because the continuous response variable (IVR) had non-normal distribution on the positive semi-axis with a significant skewness degree. The final model included sex (dichotomous variable), age (continuous variable) and severity (8). After running the full model with all interactions, variables found not to have a significant effect were removed from the model following a backwards selection criterion. The analyses were performed using R language version 2.15.2, an open-source programming language and software environment for statistical computing and graphics (www.r-project.com).

Ethics

Data on IVR evaluations were part of the STER, a study approved by the Internal Review Boards (IRBs) of L'Aquila University and all the centres participating in the study. The PK study

in 10 patients with a severe FVII deficiency administered rFVIIa in a non-bleeding state was approved by the IRB of the University Hospital in Bratislava and by the IRB of L'Aquila University (22/11/2011; comitato.etico@asl-laquila.it).

Results

IVR evaluations

The IVR distribution following rFVIIa administration in the STER cohort showed a strong positive asymmetry, with values ranging from 0.03 to 2.1 U dl-1/U kg^{-1} (mean: 0.59; median: 0.46; SD: 0.5). When subjects were grouped according to clinical severity, for major bleeders, significantly higher (p=0.04) IVR values were observed (Fig. 1). Multivariate analysis of the IVR data, which did not show an effect of age (p=0.17) or sex (p=0.41), indicated a significant influence of clinical severity.

PK evaluation

Data pertaining to PK parameters from the 10 non-bleeding patients with severe FVII deficiency are reported in Table 2. With reference to the dose-independent parameters, the PK data obtained using the FVII:C assay were significantly different from those obtained using the PTr assay (Table 2). A very good correlation of parameters over time was observed for both assays (Table 2); however, the decay of the 1/PTr curve was less biphasic than that of FVII:C (Fig. 2). Mean residence time (MRT), half-life and terminal half-life of the 1/PTr values were higher in comparison with the corresponding parameters obtained using FVII:C.

Discussion

Here we report the PK of rFVIIa in a large number of patients with severe FVII deficiency, representative of the whole clinical spectrum of FVII deficiency, with the majority of the cohort

(66.1%) accounting for symptomatic individuals and more than 20% with severe phenotypes (Table 1).

We performed extensive IVR evaluations but despite attempts to reduce the IVR variability by restricting the analysis to assays performed with thromboplastins of similar sensitivity (ISI≅1), excluding inhibitor patients and further restricting the cohort to patients with FVII:C levels ≤26%, substantial variation still remained. This finding could not be explained by sex or age; in the multivariate analysis, the only significant determinant of IVR variability was disease severity, which somehow reflected residual FVII expressed by the mutated gene. It is tempting to speculate that the presence of native FVII molecules may influence IVR. All in all, we believe that the low rFVIIa recovery and its variability may mostly be accounted for by a large extravascular distribution volume of rFVIIa, a supposition supported by the finding that the rFVIIa volume of distribution at equilibrium (Vss) was approximately 40 times that of the normal plasma volume (Table 2).

The full PK analysis preformed provided other relevant information including a low rFVIIa area under the curve (AUC), short half-life and fast clearance. In this respect it is worth noting that the terminal-phase half-life of rFVIIa appeared longer than what the MRT or half-life might have indicated. This finding, together with a prolonged pharmacodynamic effect, may be explained by a reduction in factor flow from the plasma pool to the extravascular space at the end of the decay curve, as indicated by the apparent volume of distribution, based on the terminal phase (V_z), being higher than the V_{ss}.

Mathijssen et al (12) described three cases of very severe FVII deficiency treated with rFVIIa prophylaxis in whom a very fast clearance together with very low AUC and $t_{1/2}$ (0.5–1 hour) were

observed. Of note, prophylaxis was judged applicable, safe and clinically efficacious (12). The observations of this case report were considerably strengthened by our own experience with a large number of patients, which showed that prophylaxis schedules based on an average rFVIIa total dose of 100 μg kg^{-1} per week split into three administrations were efficacious (14). Such replacement regimens may be regarded as insufficient based on the PK data reported here and in the literature. A possible explanation for the efficacy of prophylaxis may be related to the binding of FVIIa to the perivascular sites (22) from where it would be slowly released. Another possibility is that excess rFVIIa may bind to the endothelial protein C receptor on undamaged endothelial cells (23, 24); a morphological study confirmed the occurrence of rapid binding of FVIIa to the endothelium and transfer into the extravascular space, where it is likely to bind to tissue factor (TF) on the pericytes (25). Through these mechanisms, rFVIIa could remain protected from degradation and, eventually, trigger further generation of thrombin. These cell-based studies provide strong support for our main findings, namely the very large volume of distribution, low IVR and residual clotting activity when FVII:C is no longer detectable (Fig. 2). In the recent PK study conducted by Mathijssen et al. (10) a large volume of distribution for rFVIIa was also reported and the authors suggested that this finding could explain the prolonged prophylaxis effect and here we provide some clotting-related evidence to this hypothesis.

Mathijssen and colleagues also found that the FVII antigen circulated for a longer time than FVIIa (12) a finding confirmed by their more recent study on PK in FVII deficiency (10). As FVII antigen does not account for clotting activity, this introduces the question related to what is the most convenient assay for evaluating the PK features of rFVIIa. This issue was also addressed by Scharling et al. (26), who compared FVII:C with a FVIIa functional assay, showing that the latter was characterised by less variable PK features and more reliable results, especially when assessing low FVII activity levels (<2–3%). To further contribute to this clinically relevant issue, we compared the kinetics of FVII:C to that of PTr. Data were highly comparable (Table 2) with

data obtained from both assays displaying a very good correlation with time (1/PTr 'r' = -0.98 and FVII:C 'r' = -0.99). Using the PTr, PK parameters were characterised by a very slow clearance (about one-quarter of that obtained using FVII:C), a four-times larger AUC and a much longer $t_{1/2}$ (Fig. 2), differences that were all highly significant (all $p<0.001$). However, there are methodological aspects important to consider when interpreting these results; while the PTr examines an individual's TF pathway as a whole, including the patient's accelerators and inhibitors, the FVII:C assay largely depends on exogenous factors and co-factors provided by the FVII-deficient plasma, very prevalent (at least 10 times) in the assay. This may account for the very different PK results obtained using the two methods, and suggests a potential role for soluble plasma factors. Hence, the FVII:C-based findings may only partially represent the *in vivo* pro-coagulant potential and therefore provide a poor prediction of efficacy.

A limitation of this study is that the protocol used was limited to a 24-hour observation period, and therefore we did not record the tail of the PTr curve (Fig. 2). In fact, more than 20% of the 'clotting potential' examined using the PTr was still present 24 hours after rFVIIa infusion, when FVII:C was no longer detectable. The prolonged clotting correction provided by rFVIIa (as detected by the PTr) is in agreement with the observation by Brummel Ziedins (27).

In conclusion, our data support previous convincing clinical data (11-14) published on the efficacy of prophylaxis in FVII deficiency, and provides evidence for a large volume of distribution of rFVIIa and the persistence of a clotting potential when FVII:C is no longer detectable. The rapid and substantial diffusion of rFVIIa towards the extravascular spaces would trigger a yet unexplored mechanism to provide pro-coagulant activity for a longer time than expected.

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Disclosures

MM has acted as a paid consultant to Bayer, Baxter, Novo Nordisk and Pfizer advisory boards, and has received fees as an invited speaker from Bayer, Novo Nordisk, CSL Behring Symposia and Octapharma. AB has no conflicts of interest to declare. GM has acted as a consultant to Novo Nordisk and has participated in workshops and, as coordinator of the International FVII Study Group and the STER, collected financial support from Institutional Research Organizations and unrestricted funding from Novo Nordisk and charities, administered by the Internal Medicine Department of the University of L'Aquila. UM has participated in Novo Nordisk advisory board meetings and is a Novo Nordisk shareholder (purchased from his own savings). GA has acted as a paid consultant for Novo Nordisk, has also received from Novo Nordisk funding for clinical research, travel support to attend congresses and symposia and honoraria for talks. JI has acted as a speaker at training courses for Novo Nordisk, and his institution has received a grant for serving as a central laboratory on behalf of the STER. J-FS has participated in meetings sponsored by Novo Nordisk. GDiM has served on advisory boards and received honoraria and grants from Pfizer, Bayer, Novo Nordisk, CSL Behring, Boehringer and Biotest for studies unrelated to the present study. AD, CF, MG-B, FB, MP and MN have no conflicts of interest to declare.

References

1. Björkman S, Berntorp E. Pharmacokinetics. In: Textbook of Hemophilia. Wiley-Blackwell 2010; pp. 104-110.
2. Collins PW, Blanchette VS, Fischer K, et al. Break-through bleeding in relation to predicted factor VIII levels in patients receiving prophylactic treatment for severe hemophilia A. J Thromb Haemost 2009; 7: 413-420.
3. Collins PW, Björkman S, Fischer K, et al. Factor VIII requirement to maintain a target plasma level in the prophylactic treatment of severe hemophilia A: influences of variance in pharmacokinetics and treatment regimens. J Thromb Haemost 2010; 8: 269-275.
4. Mannucci PM, Duga S, Peyvandi F. Recessively inherited coagulation disorders. Blood 2004; 104: 1243-1252.
5. Mariani G, Bernardi F. Factor VII deficiency. Semin Thromb Hemost 2009; 35: 400-406.
6. Perry DJ. Factor VII deficiency. Br J Haematol 2002; 118: 689-700.
7. Mariani G, Herrmann FH, Dolce A, et al. Clinical phenotypes and factor VII genotype in congenital factor VII deficiency. Thromb Haemost 2005; 93: 481-487.
8. Di Minno MN, Dolce A, Mariani G, et al. Bleeding symptoms at disease presentation and prediction of ensuing bleeding in inherited FVII deficiency. Thromb Haemost 2013; 109: 1051-1059.

9. Berrettini M, Mariani G, Schiavoni M, et al. Pharmacokinetic evaluation of recombinant, activated factor VII in patients with inherited factor VII deficiency. Haematologica 2001; 86: 640-645.

10. Mathijssen NCJ, Masereeuw R, Holme PA, et al. Increased volume of distribution for recombinant activated factor VII and longer plasma-derived factor VII half-life may explain their long lasting prophylactic effect. Thromb Res 2013; 132: 256-262.

11. Auerswald G, Michaels L, Altisent C, et al. Prophylaxis with Novoseven® in factor VII deficiency: preliminary results from STER (Seven Treatment Evaluation Registry). Haemophilia 2010; 16 (Suppl 4): 1-158. 29P17.

12. Mathijssen NC, Masereeuw R, Verbeek K, et al. Prophylactic effect of recombinant factor VIIa in factor VII deficient patients. Br J Haematol 2004; 125: 494-499.

13. Tcheng WY, Donkin J, Konzal S, et al. Recombinant factor VIIa prophylaxis in a patient with severe congenital factor VII deficiency. Haemophilia 2004; 10: 295-298.

14. Napolitano M, Giansily-Blaizot M, Dolce A, et al. Prophylaxis in congenital factor VII deficiency: indications, efficacy and safety. Results from the Seven Treatment Evaluation Registry (STER). Haematologica 2013; 98: 538-544.

15. Mariani G, Liberti G, D'Angelo T, et al. Factor VII activity and antigen. In: Laboratory Techniques in Thrombosis. A Manual. Kluwer Academic Publishers 1999; pp. 99-106.

16. Gibaldi M. Estimation of the pharmacokinetic parameters of the two-compartment open model from post-infusion plasma concentration data. J Pharm Sci 1969; 58: 1133-1135.

17. Longo G, Matucci M, Messori A, et al. Pharmacokinetics of a new heat-treated concentrate of factor VIII estimated by model-independent methods. Thromb Res 1986; 42: 471-476.

18. Matucci M, Messori A, Donati-Cori G, et al. Kinetic evaluation of four Factor VIII concentrates by model-independent methods. Scand J Haematol 1985; 34: 22-28.

19. Bernardi F, Marchetti G, Pinotti M, et al. Factor VII gene polymorphisms contribute about one third of the factor VII level variation in plasma. Arterioscler Thromb Vasc Biol 1996; 16: 72-76.

20. Bernardi F, Arcieri P, Bertina RM, et al. Contribution of factor VII genotype to activated FVII levels. Differences in genotype frequencies between northern and southern European populations. Arterioscler Thromb Vasc Biol 1997; 17: 2548-2553.

21. McCullagh P, Nelder JA. Generalized Linear Models. 2nd ed. Boca Raton, Chapman & Hall/CRC; 1989.

22. Hoffman M, Colina CM, McDonald AG, et al. Tissue factor around dermal vessels has bound factor VII in the absence of injury. J Thromb Haemost 2007; 5: 1403-1408.

23. Nayak RC, Sen P, Ghosh S, et al. Endothelial cell protein C receptor cellular localization and trafficking: potential functional implications. Blood 2009; 114: 1974-1986.

24. Sen P, Gopalakrishnan R, Kothari H, et al. Factor VIIa bound to endothelial cell protein C receptor activates protease activated receptor-1 and mediates cell signaling and barrier protection. Blood 2011; 117: 3199-3208.

25. Gopalakrishnan R, Hedner U, Ghosh S, et al. Bio-distribution of pharmacologically administered recombinant factor VIIa (rFVIIa). J Thromb Haemost 2010; 8: 301-310.

26. Scharling B, Nielsen GG, Klitgaard T, et al. Comparison of coagulant activity of factor VII and activated factor VII activity assays when used for determination of recombinant activated factor VII levels in plasma. Blood Coagul Fibrinolysis 2007; 18: 677-684.

27. Brummel Ziedins K, Rivard GE, Pouliot RL, et al. Factor VIIa replacement therapy in factor VII deficiency. J Thromb Haemost 2004; 2: 1735-1744.

Legends to Figures and Tables

Figure 1: Comparison between IVR obtained in individuals with different clinical phenotypes.

Figure 2: Comparison between the FVII plasma concentration following the infusion of rFVIIa in patients with severe FVII deficiency (n=10) using data obtained from either the FVII:C or PTr. The solid lines represent the means, and the dashed lines represent the variance (1 SD). **Comparison between IVR obtained in individuals with different clinical phenotypes.**

Table 1: Demographic and clinical data (STER cohort)

Table 2: Kinetic parameters calculated using FVII:C levels (N=10) and 1/PTR (mean values only of dose-independent parameters)

Table 1. Demographic and clinical data (STER cohort)

Demographic data (N=56)	
Age (years)	
Mean (SD), median (range)	32.8 (24.2), 23 (2–81)
Weight (kg)	
Mean (SD), median (range)	58.3 (23.7), 60 (11–110)
FVII:C level (%)	
Mean (SD), median (range)	7.6 (8.6), 4 (<1–26)
Sex (F/M)	32/24
Asymptomatic, n (%)	19 (33.9)
Symptomatic, n (%)	37 (66.1)
Major bleeders	12 (21.4)
Minor bleeders	25 (44.6)
Clinical data	
Bleeding type, frequency (%)	
Menorrhagia	33.90
Epistaxis	32.10
Gum bleeding	28.60
Bruising	28.60
Muscle haematoma and subcutaneous haematoma	28.60
Haemarthrosis	17.90
Other symptoms	17.90
Haemoperitoneum	12.50
Post-operative bleeding	8.90
Haematuria	7.10

Haemorrhoidal bleeding	7.10
Gastrointestinal bleeding	3.60
Central nervous system bleeding	1.80

Table 2. Kinetic parameters calculated using FVII:C levels and 1/PTR (mean values only of dose-independent parameters)

	FVII:C				1/PTr	
Parameter	**Mean**	**SD**	**Min**	**Max**	**Mean**	**p**
Correlation XY (r)	−0.99	0.01	−1.00	−0.98	−0.98	
Number of points λ z	3.80	1.32	3.00	6.00	6.22	<0.001
Lambda (λ) z (1/hours)	0.20	0.07	0.06	0.33	0.06	<0.001
HL λ z (hours)	4.23	2.58	2.10	11.15	13.81	<0.001
AUC_{last} (U*h $dl^{-1)}$	218.84	27.46	183.18	267.55	-----	
V_{zobs} (dl $kg^{-1)}$	24.98	8.86	14.48	42.12	------	
CL_{obs} (dl h $kg^{-1)}$	4.41	0.72	2.62	5.07	------	
$AUMC_{last}$ (U*h^2 $l^{-1)}$	681.87	252.20	494.37	1324.16	------	
MRT_{last} (hours)	3.05	0.72	2.37	4.95	8.38	<0. 001
V_{ssobs} (dl $kg^{-1)}$	14.72	3.83	11.36	25.02	------	
$t_{1/2}$ (hours)	2.12	0.50	1.64	3.44	5.80	<0.001
I-IVR (U dl^{-1}/U kg^{-1})	0.57	0.28	0.12	1.02	------	
P-IVR (%)	25.33	12.47	5.4	45	------	

AUC_{last}, area under the plasma concentration–time curve from time zero to the last measurable concentration; $AUMC_{last}$, area under the moment curve from time zero to the last measurable concentration; CL_{obs}, clearance; HL λ z, terminal phase half-life; I-IVR, incremental *in vivo* recovery; lambda (λ) z, terminal elimination rate constant; MRT_{last}, mean residence time from time zero to the last measurable concentration; $t_{1/2}$, distribution phase half-life; V_{ssobs}, apparent volume of distribution at equilibrium; P-IVR, percentage IVR; V_{zobs}, apparent volume of distribution during the terminal phase.

Extra Table

What is known on this topic:

- rFVIIa has a very short half-life and rapid clearance
- rFVIIa rapidly diffuses towards the extravascular spaces
- rFVIIa is effectively used for prophylaxis in FVII deficiency

What this paper adds:

- rFVIIa has a low ex vivo Recovery
- Ex vivo recovery is independent of age and sex
- Ex vivo recovery is higher in major bleeders
- rFVIIa has a very large volume of distribution at the steady state
- The PTr-related kinetic data demonstrate that there is a considerable clotting potential available when FVII:C levels are no longer detected in the bloodstream

Figure 1.

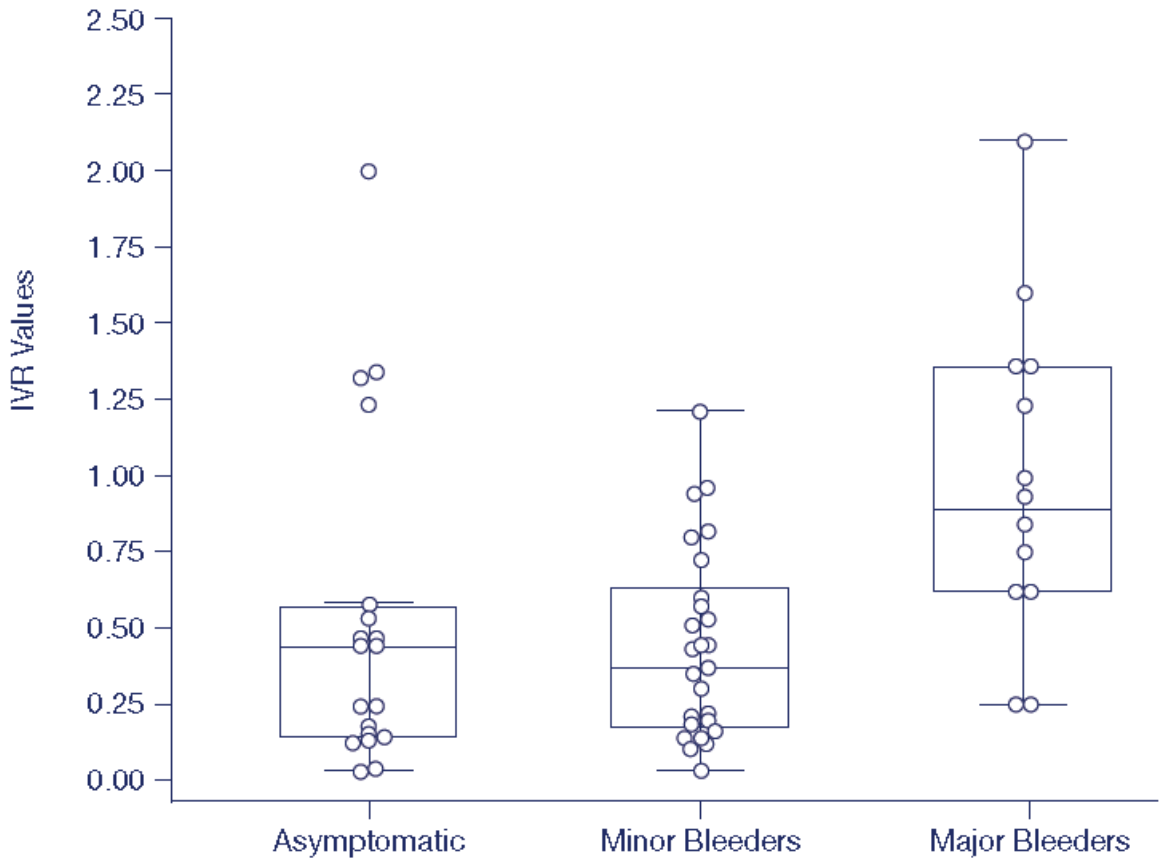
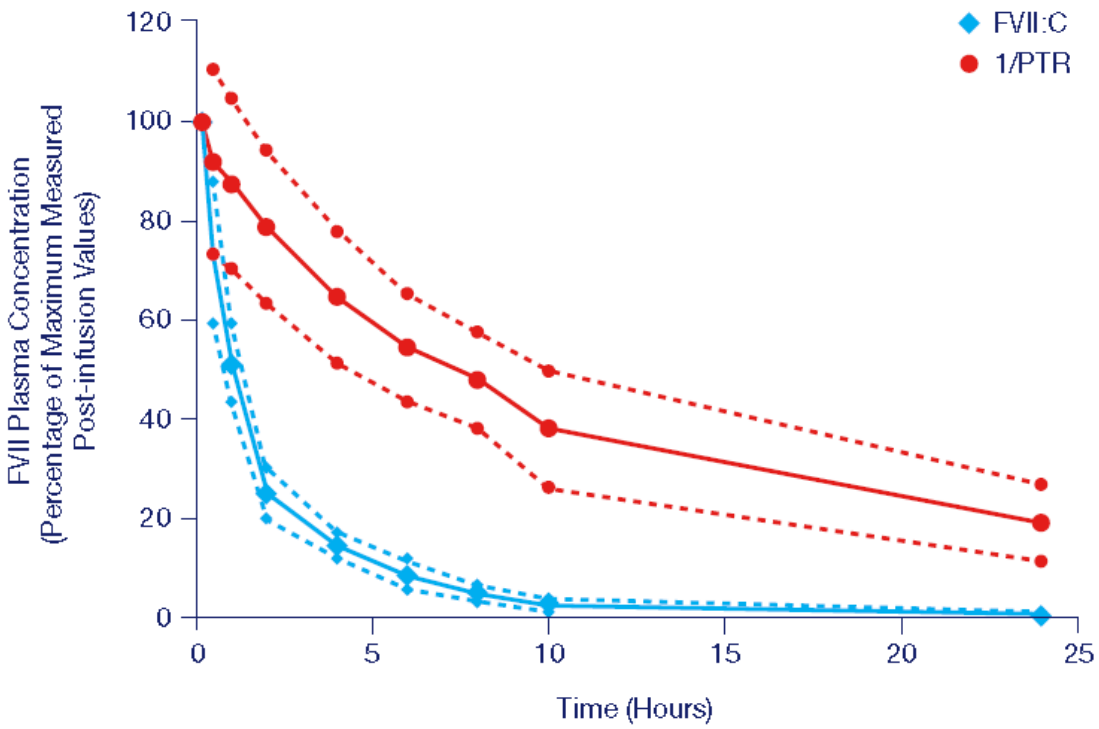

2.50
2.25
2.00
1.75
1.50
1.25
1.00
0.75
0.50
0.25
0.00
IVR Values
Asymptomatic
Minor Bleeders
Major Bleeders

Figure 2.


FVII:C
1/PTR
FVII Plasma Concentration
(Percentage of Maximum Measured
Post-infusion Values)
120
100
80
60
40
20
0
0
5
10
15
20
25
Time (Hours)